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All correspondence regarding articles from the Department of Agriculture should be addressed to Karl F. Kellerman, Journal of Agricultural Research, Washington, D. C.

All correspondence regarding articles from State Experiment Stations should be addressed to J. G. [illegible]an, New Jersey Agricultural Experiment Station, New Brunswick, N. J.

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ERRATA AND AUTHORS' EMENDATIONS

Pages 60–63, legends to figures 3–8, "phosphate" should read "phosphorus."

Page 175, lines 45–46, Mr. Ainslie informs the author that he has never succeeded in rearing *Pyrausta ainsliei* to maturity on *Nelumbo lutea* as here stated. In the discussion of *P. ainsliei* it should be stated that the old name *P. obumbratalis* Lederer may eventually prove to apply to this species. Without an examination of the genitalia of Lederer's types in Europe, however, it can not be determined whether his name applies to this species or to *P. penitalis* Grote, if to either. In the meantime it is safest to employ a definite name to a definite concept.

Page 333, Table III, column 8, next to last line, and column 9, next to last line, "3.329" should read "3.320."

Page 335, Table VI, "phosphates" should read "phosphate."

Page 339, Table XIII, column 5, line 8, "92" should read "80."

Page 393, line 27, "in part by weight" should read "in parts by weight."

Page 394, Table I, column 9, line 10, "54.5" should read "5.45." Note *a*, "expressed in parts" should read "expressed in parts by weight."

Page 395, Table II, continued, column 6, line 4, "1 : 1.83" should read "1 : 0.83." Second part of table, column 6, lines 1 and 2, "1 : 1.05" and "1 : 1.37" should be transposed. Line 10 of text, "basal ratio" should read "basal ration."

Page 445, line 15 of text, "from 13.3 larvæ to each fruit in 1916 to 20.3 in 1917 and to 34.6 in 1918" should read "from 13.3 per cent in 1916 to 20.3 per cent in 1917 and to 34.6 per cent in 1918."

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NOTES ON THE COMPOSITION OF THE SORGHUM PLANT[1]

By J. J. WILLAMAN, *Plant Chemist*, R. M. WEST, *formerly Assistant Chemist*, D. O. SPRIESTERSBACH, *formerly Research Assistant*, and G. E. HOLM, *Research Assistant, Division of Agricultural Biochemistry, Minnesota Agricultural Experiment Station*

I.—INTRODUCTION

Since 1877, when the United States Department of Agriculture undertook the investigation of sorghum (*Sorghum vulgare*) as a source of crystallized sugar, many thousands of analyses of sorghum juice, from many different varieties, have been made and published. As a result, there is a well-established fund of knowledge concerning the kinds and quantities of sugars in the juice, especially for the more temperate regions of the United States. Considerable work has been done in this and in other countries on the effect of removing the seed heads on the composition of the juice. Also a little work has been done on the practices followed in the manufacture of sorghum sirup. However, when one of the present writers, R. M. West, undertook in 1912 to place the sorghum industry in Minnesota on a better economic and scientific basis, the need for further chemical investigations was seen at once. It was apparent (1) that, considering the effect of climatic factors on the composition of the cane, more exact knowledge was needed concerning the behavior of sorghum grown in the most northern limit of its range; (2) that the utilization of the cane somewhat prior to maturity, and very often after being killed by frost, would be necessary in order to lengthen the milling season as much as possible; (3) that the methods of defecation and evaporation in vogue were decidedly in need of improvement and standardization; (4) that for economic reasons the small-scale manufacture of sorghum sirup, with inefficient mills, little or no defecation, and slow boiling, would have to give way to large-scale production or the rapid decrease in production of sirup, as witnessed for the last thirty years, would no doubt continue. The investigations at this Station resulted in the accumulation of considerable data of both scientific and practical interest. The

[1] Published with the approval of the Director as Paper 170, Journal Series, Minnesota Agricultural Experiment Station.

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latter have been compiled in a Station bulletin;[1] the former are set forth in the present paper.

II.—METHOD OF INVESTIGATION

VARIETY TESTS.—Several varieties of cane grown on University Farm, St. Paul, were tested for several consecutive years to ascertain their behavior in this region.[2] Analytical studies were made on but three varieties, however—Minnesota Early Amber, an old, very well-established, and almost universally grown variety not only in Minnesota but in the whole country; Early Rose, a variety isolated from Early Amber in southern Minnesota about 15 years ago; and Dakota Amber, another selection from Early Amber that was made in South Dakota. The data in this paper are the averaged results from these three varieties.

SAMPLES.—The data on the progressive changes in composition of the cane cover a period of three growing seasons. Not all data, however, were obtained for all the years. The stages of growth at which samples were taken were as follows:

(1) When the panicles first appeared.

(2) When the panicles were wholly emerged.

(3) When the anthers of the blossoms appeared at the middle of the panicles.

(4) When the panicles were in full bloom.

(5) When the seed was in milk.

(6) When the seed was in soft dough.

(7) When the seed was in hard dough.

(8) When the seed was brittle and mature.

During these stages the plants increased in height by 2 or 3 feet.

In taking samples, 6 to 10 plants were selected which were of nearly the same stage of growth and which at the same time were as representative as possible of the plot. Two samples were cut from each plot at each stage of growth. One was weighed, sacked, and dried; the other was weighed, stripped, topped, and the juice extracted by pressing with a small 3-roll power mill.

ANALYSIS.—The leaves, tops, cane, juice, and bagasse were weighed separately and the weights recorded, together with the losses in weight during stripping and pressing. The leaves, tops, and bagasse were sacked separately and, together with the sample of the whole plant, dried as rapidly as possible in a steam oven to less than air-dry moisture content. The samples were then exposed to the air of a well-ventilated room until there was no further increase in weight due to absorption of moisture. The finely ground air-dried material was resampled and the usual proximate determinations were made, together with an analysis of the ash for

[1] WILLAMAN, J. J., WEST, R. M., and BULL, C. P. SORGHUM AND SORGHUM-SIRUP MANUFACTURE. Minn. Agr. Exp. Sta. Bul. 187.

[2] Acknowledgments are due to Prof. C. P. Bull, of this Station, for the agronomic phases of this work.

the percentage of potash and phosphoric acid. For these determinations the official methods (*20*) [1] were followed, except that crude fiber was determined by the modified Sweeney (*10*) method, and the preparation of the sample for the potash and phosphoric acid was accomplished by the modified wet ignition method (*18*).

III.—PROPORTION OF LEAVES AND TOPS TO CANES

Since cane is brought to the mills in various conditions, such as fresh whole cane with or without seed heads or leaves or both, and partially dried cane with or without tops and leaves, it is desirable that the average proportion of these three parts be known by the mill operator in computing the value of the various grades of cane. Table I presents the average figures for six plots in 1913.

TABLE I.—*Relative percentage of leaves, seed heads, and clean cane in whole cane when fresh and in whole cane when partially dried*

Stage of growth.	Whole cane, fresh.			Whole cane, partially dried.[1]		
	Leaves.	Tops.	Clean cane.	Leaves.	Tops.	Clean cane.
Panicles appearing	19.8	8.4	71.8	.....	.....	.....
Panicles out	17.2	8.4	74.4	.....	.....	.....
Blossoms appearing	15.8	8.6	75.6	.....	.....	.....
Full bloom	15.2	8.2	76.6	.....	.....	.....
Seed in milk	16.5	11.1	72.4	.....	.....	.....
Seed in dough	15.3	14.1	70.6	.....	.....	.....
Seed dry	16.0	16.2	67.8	10.0	11.0	79.0
Seed mature	14.9	16.7	68.4	9.0	10.0	81.0

[1] Computed from various field data.

Collier (*5, p. 142*) reports 72 per cent of clean cane from fresh material. He also says that the leaves constitute 15 per cent of the topped stalks. This figure appears rather high, for it is practically the percentage of leaves in the whole cane as shown in the above table. From the above data the writers were able to construct a table for the use of manufacturers, by which they could compute the value of a ton of cane according to its condition when weighed at the factory.[2]

IV.—PROXIMATE COMPOSITION OF THE PLANT

Many analyses are on record of the proximate constituents of the various parts of the sorghum plant, designed to show its feeding value. Most of them agree substantially with the data obtained in the present work, at least as regards the general trend of development of the various constituents. It would be futile to review these analyses. The present data have been calculated in various ways in order to reveal facts not

[1] Reference is made by number (italic) to Literature cited, p. 30731.

[2] WILLAMAN, J. J., WEST, R. M., and BULL, C. P. *op cit.*

hitherto pointed out concerning the physiology of this plant and the relations between the various parts of the plant as maturity is approached. It is well to keep in mind, while perusing the graphs and data, that the sorghum plant, so far as we are concerned in the present investigations, is cultivated primarily for its sugar content. The data repeatedly reveal the subservience of all other constituents to the sugars. This same phenomenon has been pointed out for many other plants which specialize in the production of some one class of substances, as the starch in potato tubers and in corn seed, the sugars in fruits, the oil in flax, peanuts, and soybeans, and the sucrose in sugar beets and in sugar cane.

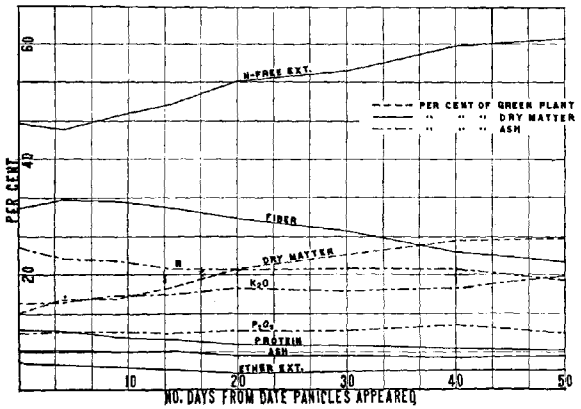

FIG. 1.—Development of the proximate and mineral constituents of the sorghum plant in the later stages of growth.

The results of the analytical studies are presented only in graphs, since their presentation in tables adds nothing to what is given in the charts.

Figure 1 shows the composition of the whole plant, expressed as percentages of the green plant and of the dry matter. The ash constituents are expressed as percentages of the total ash. The nitrogen content is computed to the ash basis, so as to make it comparable to the phosphorus and potash. There is a gradual increase in dry matter from about 12 per cent to about 26 per cent. Of this dry matter, the most prominent constituents are the fiber and the nitrogen-free extract. The latter is mostly starch, dextrose, and levulose in the younger stages, and mostly sucrose in the older. The percentages of fiber and of the other carbohydrates undergo progressive changes which are almost exactly equal to each other but opposite in character—that is, the percentage of

decrease of the fiber is the same as the percentage of increase of the soluble carbohydrates. The percentages of crude protein, ash, and ether extract remain practically the same throughout growth.

When these percentages are computed to the absolute weight of each constituent in one plant, the same facts, with but slight modification, are apparent. Figure 2 presents these data. The actual weights of protein, fat, and ash in each plant increase but slightly during the periods of growth studied; the fiber about doubles in weight, while the nitrogen-free extract trebles in weight. The stages of development studied here represent not only maturation of the plant but actual growth, the height increasing by 2 or 3 feet during these stages. It is apparent that the plant absorbs practically all of its mineral requirements, including nitrogen, during the early stages of growth, that it also lays down the necessary structures of protein and fiber during these stages, and that during the final maturation periods all the energies of the plant are directed toward the filling out of the seed and the storing of sugar in the cells of the cane. This program of development may prove to be the rule in all plants, as it has already been proved in several of them, notably in wheat, by Thatcher (*16*).

The sharp decline in the curves in figure 2 for the last growth period is explained by the fact that the plots had been culled of the larger plants and smaller plants had to be chosen for the final stage. Thus the percentage curves continue in the direction anticipated, while the curves of absolute weights show a declination.

The above observations concern the whole plant. Considering now the separate parts of the plant, it is found on examination of figure 3 that the leaves undergo changes in composition which are in many ways similar to those of the whole plant. The nitrogen-free extract exhibits a marked increase during the later stages. This is not paralleled by an equal decrease of fiber, however, as was the case with the whole plant. The fiber remains constant, both relatively and absolutely (fig. 4). The percentage of protein undergoes an appreciable decrease, while the absolute weight of it remains practically constant. The changes in dry matter are closely parallel to those of the nitrogen-free extract. This is corroborated by Collier's analyses of the juice of leaves (*5, p. 142*), which showed a considerable increase of sugars in the more mature stages. These data would indicate that the more mature the leaves the higher their feeding value.

Figures 5 and 6 present the curves for the composition of the tops. In the preparation of the samples the cane was cut off just below the lowest stem of the seed head, and the whole head used in the analyses. This of course resulted in the earlier samples' consisting mostly of stems, hence the high fiber content. Later, due to the filling out of the seeds with starch, the percentage of fiber underwent a marked decrease, while

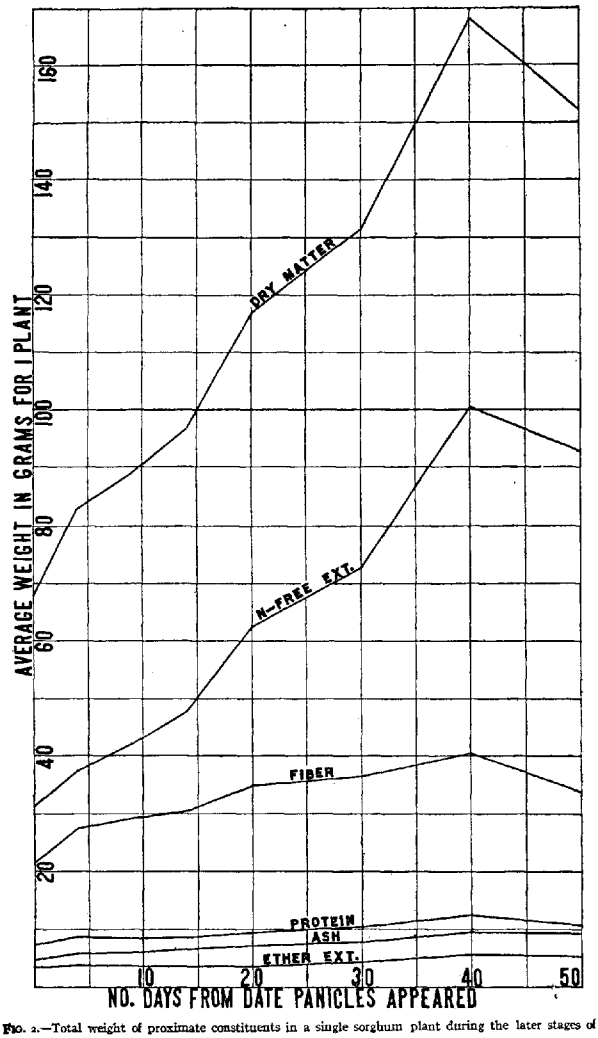

FIG. 2.—Total weight of proximate constituents in a single sorghum plant during the later stages of growth.

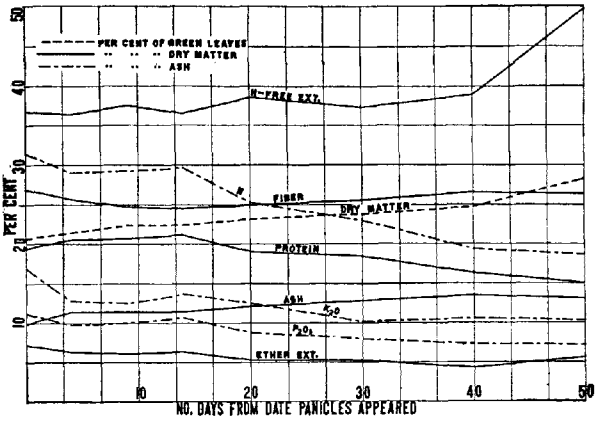

FIG. 3.—Development of the proximate and mineral constituents of the leaves of sorghum.

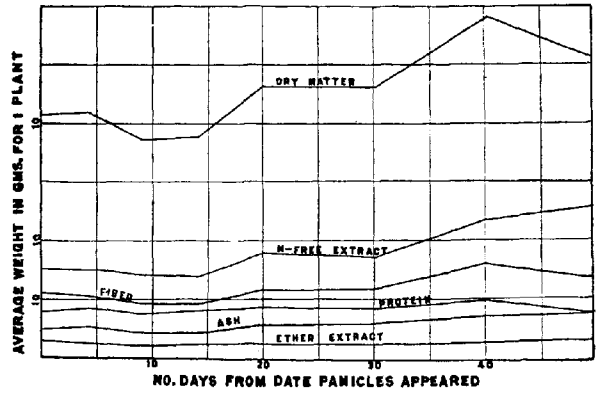

FIG. 4.—Total weight of proximate constituents in the leaves of a single sorghum plant.

its absolute amount remained nearly constant. The dry matter and the nitrogen-free extract again parallelled each other, while the protein, ash, and ether extract remained rather constant.

Figures 7 and 8 contain graphs for the composition of the bagasse. Great reliance can not be placed upon these data, since the amount of juice obtained from the cane varied from 34.5 to 37 per cent of the weight of the cane. The bagasse as analyzed thus contained a considerable proportion of juice. The relative proportions of the various constituents are similar to those of the leaves and tops.

Some interesting relations can be seen by collecting the percentage curves for individual constituents for various parts of the plant on the

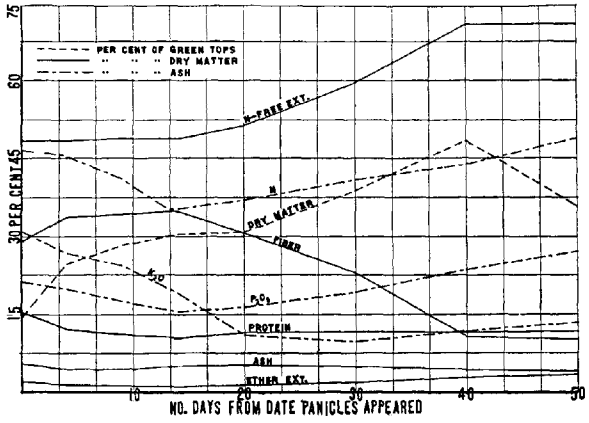

FIG. 5.—Development of the proximate and mineral constituents of the seed heads of sorghum.

same chart. In this way the history of each constituent can be viewed in its relation to the whole plant.

Figure 9 contains the dry matter curves. As would be expected, there is a general increase in the percentage of dry matter throughout the whole plant. The most marked increase is in the tops. The apparent abnormality of a sharp decrease in the last period of this curve is unexplained; it is no doubt an analytical error. A large portion of the increase in dry matter of the bagasse is probably due to the sugars of the retained juice, since the two curves parallel each other very closely, and since only about two-fifths of the juice was expressed by the small experimental mill.

Figure 10 contains the curves for the crude protein. As has been pointed out above, there is not only a regular decline in the percentage

of protein throughout the plant but the absolute weight of protein in each plant remains practically constant through the stages of growth studied here. This is no doubt brought about by the great increase in nitrogen-free extract sugars in the cane and starch in the seed heads.

Figure 11 contains the ether extract curves. They indicate that in all parts of the plant except the tops the ether extract remains constant. In the tops there is a slight increase. This constituent is not prominent

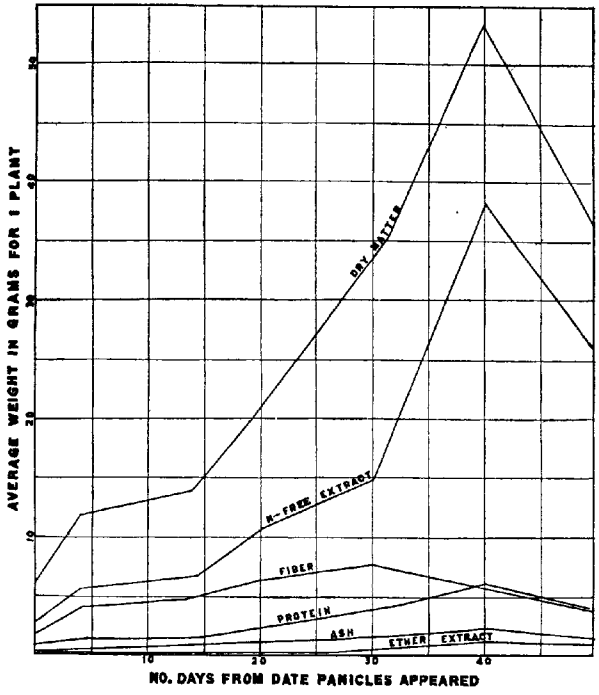

FIG. 6.—Total weight of proximate constituents in the seed head of a single sorghum plant.

in any part of the plant. The higher percentages in the leaves are due in large part to the chlorophyll.

The crude fiber curves are shown in figure 12. The curves for the bagasse and for the leaves are practically horizontal. The very sharp decrease in the fiber in the tops causes a marked decrease in the percentage of fiber in the whole plant also.

The nitrogen-free extract is the proximate constituent most characteristic of the sorghum plant and the constituent for which the plant is grown. The curves for this extract in the various parts of the plant are assembled in figure 13. There is a pronounced increase in all parts, most noticeable in the tops. Although the percentage of increase of nitrogen-free extract is greatest in the tops, the absolute increase is almost equally great in the juice because of the accumulation of sugars. This fact is brought out in figures 5, 7, and 8.

The ash curves are given in figure 14. There is a very apparent tendency for the mineral material to accumulate in the leaves. This is shown not only on the percentage basis but also on the basis of the absolute weights of ash per plant (figs. 6, 7, and 8). Figure 15 indicates

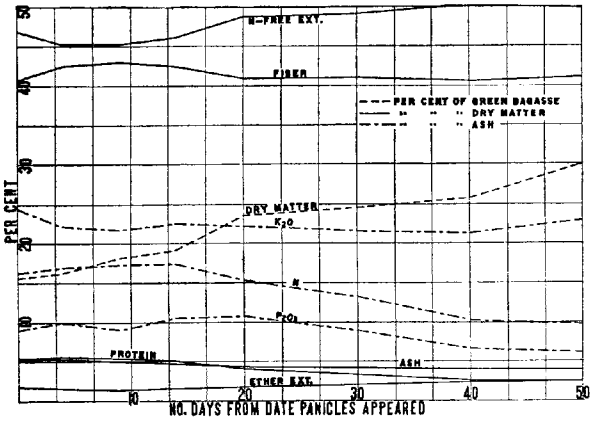

FIG. 7.—Development of the proximate and mineral constituents of the bagasse of sorghum.

that this accumulation of ash in the leaves is not due to potassium or phosphorus, for the percentage of these undergoes a marked decrease. No doubt calcium and silicon would be found responsible for the increase in mineral matter if analyses had been made for these elements. The mineral matter in the other parts of the plant remains practically constant throughout the periods of growth studied here. Figure 15 shows that potassium is more abundant than phosphorus in all parts of the plant except the tops, where the phosphorus towards maturity accumulates in greater amount. This relation is perfectly normal; it obtains in the seed of practically all plants. The prominence of nitrogen in the tops and in the leaves, and of potassium in the stalks (bagasse), is also characteristic of most plants.

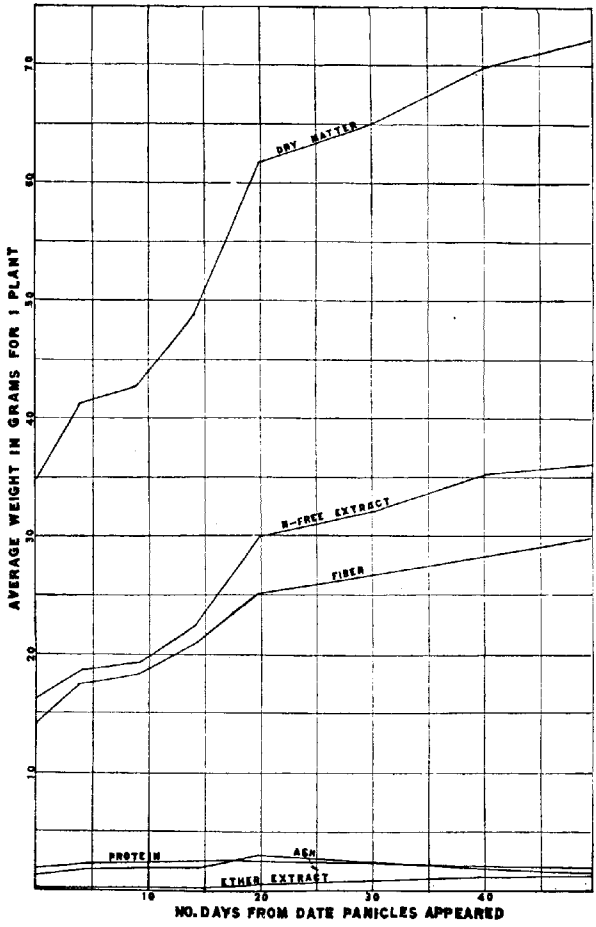

FIG. 8.—Total weight of proximate constituents in the bagasse of a single sorghum plant.

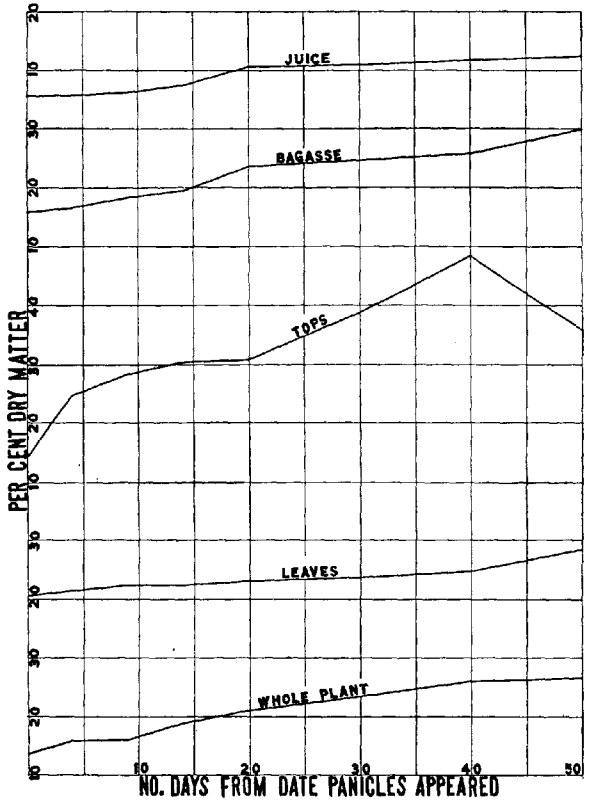

FIG. 9.—Development of the percentages of dry matter in the various parts of the sorghum plant.

V.—COMPOSITION OF THE JUICE

The juice of sorghum has naturally received greater attention at the hands of analysts than any other portion of the plant. Since, however, in practically all the previous work on sorghum only sucrose, reducing sugars, and solids-not-sugar were determined, it was thought desirable to make a more thorough investigation and attempt to acquire information concerning (1) the kinds of carbohydrates present, (2) the character of the

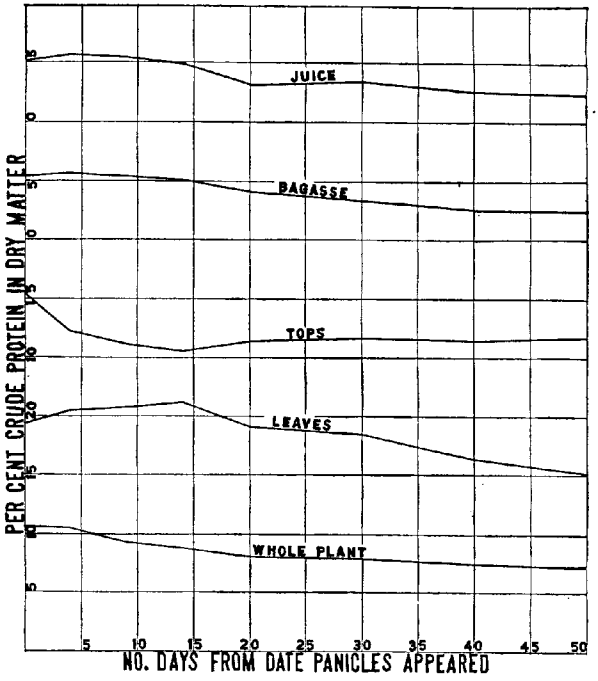

FIG. 10.—Development of the crude protein in the various parts of the sorghum plant.

noncarbohydrate solids, (3) the distribution of sugars in the cane, (4) the history of these various constituents during the growth of the plant, and (5) the effect of the removal of the seed heads on the sugar content of the juice.

(1) KINDS OF CARBOHYDRATES PRESENT.—Dextrose, levulose, and sucrose are the only sugars definitely identified. Raffinose could not be detected by the mucic acid reaction for galactose, nor maltose by the

osazone reaction. Sugar cane has also failed to yield these two sugars. Starch and gums are known to be present; but the latter have not been identified heretofore, although they are held responsible for the failure of sorghum as a source of crystallized sugar. In the sugar cane, Maxwell (*11*) found that the "so-called gums" consist largely of pentosans and hexosans. The pentosans are considered to be mostly xylan.

In the investigations reported here, the ordinary quantitative determinations of sucrose, dextrose, and levulose were also taken as means of identification when maltose and raffinose had been proved absent. Sucrose was determined by the Clerget method, and dextrose and levulose by the formula given by Wiley (*21*, *p. 360*). The discussion of these sugars will be given in subsections 3 and 4 of this section.

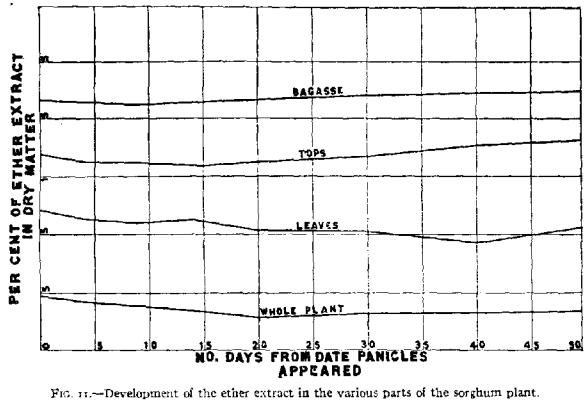

FIG. 11.—Development of the ether extract in the various parts of the sorghum plant.

The investigation of the gums and the nonsugar constituents of the juice was made the subject of a thesis by one of the writers, G. E. Holm. A barrel of juice was obtained from a sorghum factory and preserved by freezing. To a certain degree the juice was concentrated by the freezing, so that the lower portions had a slightly higher specific gravity. This was taken into account, however, in the subsequent analyses. To prove that the freezing did not alter the amount of material precipitated by an equal volume of 95 per cent alcohol, 1,000 cc. of juice were divided into two parts. One part was frozen, then thawed, and then both portions precipitated with alcohol. The unfrozen portion yielded 1.4180 gm. dry precipitate, the frozen portion 1.3940 gm.

That portion of the alcoholic precipitate which would not redissolve in boiling water was subjected to hydrolysis with dilute sulphuric acid for 20 hours. It was cooled and filtered, and then the filtrate subjected to

the tests described above for galactose, arabinose, and xylose, with the following results:

Test for xylose	+
Mucic acid for galactose	−
Osazones for galactose	−
Osazones for arabinose	−

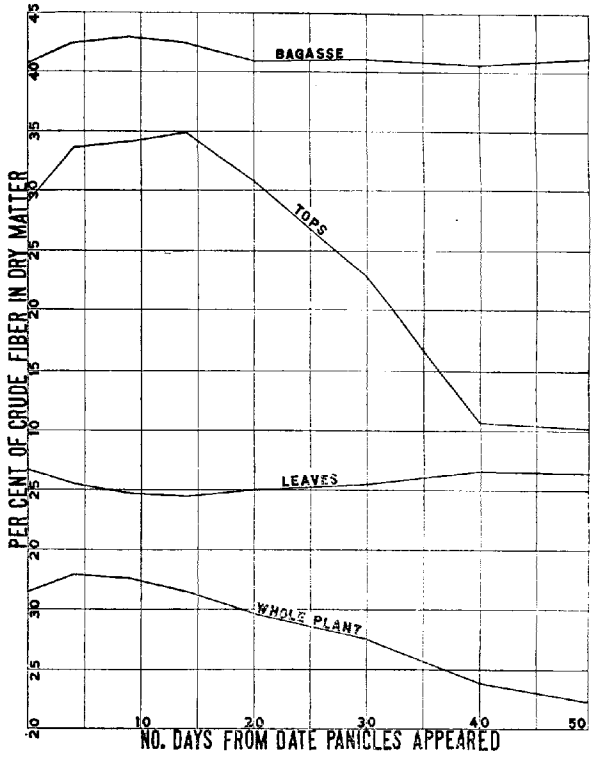

FIG. 12.—Development of the crude fiber in the various parts of the sorghum plant.

This indicates that the carbohydrate contained in the insoluble portion of the alcoholic precipitate is xylan, or at least that xylose-bearing cellular material is included in it.

The conclusion can be drawn from the above results that the material precipitated by equal volumes of alcohol from sorghum juice consists of protein, cellular material that was in suspension in the juice, and gums.

Starch is always found in sorghum juice. It will be discussed in subsection 4 of this section.

(2) THE NONSUGAR SOLIDS.—This classification will include both nonnitrogenous and nitrogenous compounds. In the former group the organic acids are the most prominent. Malic acid is usually considered

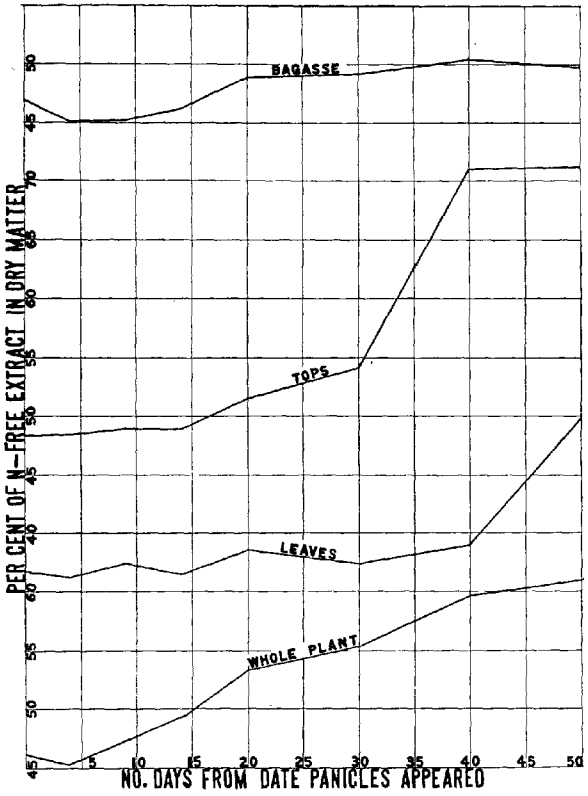

FIG. 13.—Development of the nitrogen-free extract in the various parts of the sorghum plant.

to be the characteristic acid of sorghum juice. Parsons (*12*), however, found the scale on the sugar pans to be about two-thirds calcium hydrogen aconitate. In a series of unpublished experiments by the writers, malic, citric, and tartaric acids were found to be invariably present. Also

during the isolation of nitrogenous compounds described below, calcium oxalate crystals were separated and identified in appreciable quantities. Lack of opportunity has prevented the study of the history of these acids during the development of the plant. Titration data on juices are not included in this paper. They bear little significance, since in all plant juices the acids occur as salts to a considerable degree and the titration gives no idea of the absolute quantity of acids present. Suffice it to say at this place that malic, tartaric, oxalic, citric, and aconitic acids are present in sorghum juice.

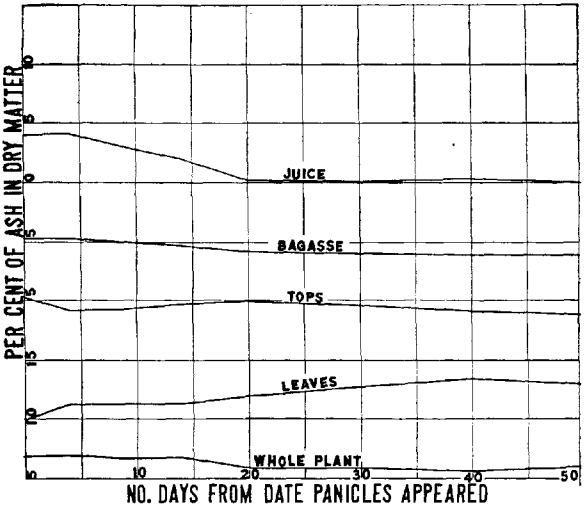

FIG. 14.—Development of the ash content in the various parts of the sorghum plant.

These data prove conclusively the presence of pentosans in the gums of sorghum juice. It is believed that the substances precipitated by alcohol are true gums and not pectins, since the jell test described by Goldthwaite (6) gave negative results. The ash, by qualitative tests, was shown to consist mostly of calcium and magnesium with some potassium. This is in accordance with the description of true gums by Haas and Hill (7, *p.* 120). It is also in accordance with the findings of Anderson (3) that sorghum juice, because of the gums present, absorbs over twice as much calcium hydrate as is accounted for by titration.

One-half liter of juice of specific gravity 1.078 was precipitated by alcohol. The precipitate was dried and weighed. A portion of it was used for ash and for nitrogen determinations. The rest was boiled in

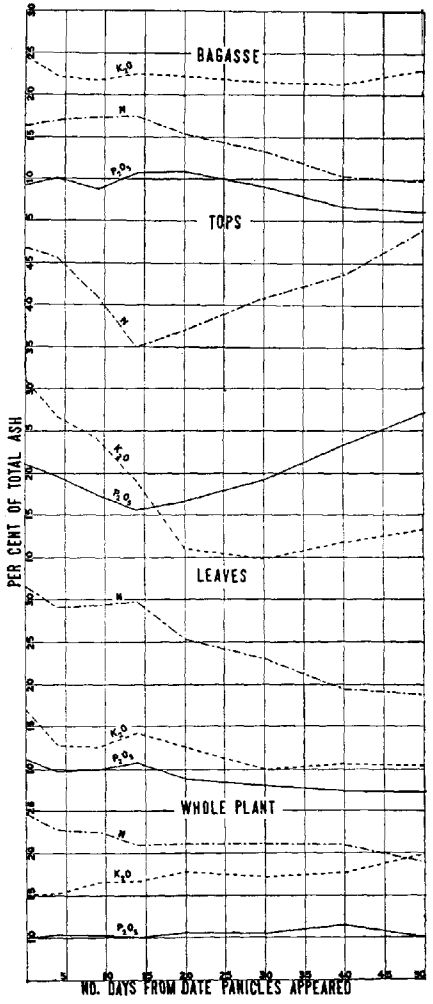

FIG. 15.—Development of the composition of the ash in various parts of the sorghum plant.

water for 15 minutes, filtered from the insoluble portion, and the soluble portion precipitated by alcohol. The results were as follows:

Total alcohol precipitate from 500 cc. juice	4.30 gm.
Percentage of juice	0.399.
Composition of precipitate:	
Proteins (N×6.25)	12.00 per cent.
Ash	22.22 per cent.
Gums (by difference)	65.28 per cent.
Solubility of precipitate:	
Insoluble in boiling water	23.87 per cent.
Soluble in boiling water, precipitated by alcohol	26.40 per cent.
Soluble in boiling water, not precipitated by alcohol	49.73 per cent.

No doubt most of the portion that does not redissolve in hot water consists of protein that was coagulated by the alcohol and of fine particles of pith and fiber that were suspended in the original juice. The high ash content is probably due to the bases that are always associated with the true gums. There is a possibility that calcium citrate constitutes a portion of it, since this salt is rather insoluble in alcohol.

That portion of the alcoholic precipitate which redissolved in water was hydrolyzed for 15 hours with $N/5$ sulphuric acid. The acid was removed with barium hydrate and the filtrate tested for xylose, arabinose, and galactose. The osazones were formed and compared with those from the pure sugars. The mucic acid test was employed for galactose, and the Bertrand reaction (*1, p. 101*) for xylose. The results are as follows:

Bertrand test for xylose	−
Mucic acid test for galactose	+
Osazone for galactose	±
Osazone for arabinose	−

These results indicate the presence of galactan in the gums. It was surprising to find no pentoses in this solution, since they should be present whether the alcoholic precipitate consists of true gums or pectic substances. In the belief that the failure to find pentoses was due to the osazone method of detection and not to their absence from the solution, they were sought for by the phloroglucid method. No more sorghum juice being available, some sorghum sirup was diluted with water, the gums precipitated and reprecipitated with alcohol, dried, weighed, and analyzed, with the following results:

Ash	19.09 per cent
Pentosan	3.76 per cent.

The nitrogenous constituents were studied in the juice preserved by freezing. The writers know of no previous work on this subject. Some rather careful work has been done with sugar-cane juice, and that is the only basis for comparison in the present instance.

Since Browne (*4*) and Maxwell (*11*) have recommended and used the ratio of protein to nonprotein nitrogen in cane juice as an indication of the stage of maturity of the plant, these determinations were made on sorghum juice by means of Stutzer's reagent (*20, p. 38*). The results for 50-cc. samples follow:

	Percentage of nitrogen in juice.	Percentage of total nitrogen as—	
		Albuminoid nitrogen.	Amid nitrogen.
Sample 1	0.0204	39.2	60.8
Sample 2	.0191	35.8	64.2

In sugar-cane juices the albuminoid nitrogen may vary from 20 to 70 per cent of the total nitrogen, depending upon the age of the juice and the method of extracting the juice. The high proportion of "amid nitrogen" in sorghum is significant from the viewpoint of sirup manufacture since it represents the impurities which are not coagulated by heat and which are not in large part removed by lime defecation. They no doubt contribute to the flavor of the sirup.

It was found that lead acetate, lead subacetate, and mercuric nitrate would each precipitate a different amount of material from the juice. Therefore these reagents were used to remove fractionally the nonsugar solids from the juice. The mercuric nitrate was added to the filtrate from the lead acetate precipitate and the lead subacetate brought down the third fraction. The following are the data for a typical example of this precipitation on a juice containing 0.0265 per cent of nitrogen:

Precipitant.	Percentage of nitrogen in juice.	Percentage of nitrogen precipitated by reagents.
Lead acetate	0.0071	27.30
Mercuric nitrate	.0063	24.23
Lead subacetate	.0062	23.84
Last filtrate	.0065	24.52
Total	.0261	99.89

Thus, three-fourths of the nitrogen can be removed by these precipitants.

Forty-eight liters of juice were treated in this way, the precipitates being separated and washed by centrifuging. The precipitates were decomposed with hydrogen sulphid, the metallic sulphid filtered off, and the filtrate concentrated *in vacuo* at 30° C. to a thin sirup. From all

three fractions brilliant octahedral crystals of calcium oxalate separated. Their crystal form, together with their solubilities,[1] established their identity as calcium oxalate. They were removed by filtration and washing and the filtrates concentrated to thick sirups. All three again produced the same kind of crystals; in this case they were spherical masses, consisting of brilliant radiating needles. The sirups were diluted with water, and the crystals filtered off and recrystallized three times. On ignition they left no ash; they dissolved in large quantities of boiling water; they decomposed at 286°–288° C. It is believed they are crystals of the "impure leucine" described by Hawk (*8*, *p. 492*) and by Abderhalden (*2*, *p. 559*), who designates them l-leucin, and states a decomposition point of 293°–295° C. Shorey (*15*) isolated a compound from Hawaiian cane juice that is no doubt identical with the present one.

The three filtrates were again concentrated to a thick sirup. That from the lead acetate fraction yielded a very small quantity of crystals of two forms—one, long prisms; the other, flat hexagons. Both were insoluble in cold water, alcohol, and dilute acetic acid, but soluble in dilute sodium-hydroxid solution. The hexagonal plates when dissolved in hot water and treated with lead acetate slowly produced a dark color, showing the possibilities of its being cystin. The amount of material available was too small to perform any further tests. The long prism-shaped crystals could not be identified, although they had the appearance of aspartic acid.

The filtrates from the last crop of crystals of the mercuric nitrate and the lead subacetate fractions, after standing for some time, deposited small quantities of wedge-shaped crystals. They were slightly soluble in water, insoluble in alcohol and in ether, decomposed at 207°–210° C. (Abderhalden states 213° C. for d-l-asparagin), were acid in reaction, liberated ammonia when treated with alkali, and gave the pyrrol test (*13*) for asparagin. These reactions indicate that the crystals were d-l-asparagin. Maxwell found both asparagin and aspartic acid in cane juices. Later, Shorey (*14*) found that glycocoll is the "principal amid" of sugar cane, and that asparagin is not present. Since our preparation liberated ammonia with alkalies, it is no doubt not glycocoll. Attempts to isolate the latter have failed.

The filtrates were neutralized with calcium carbonate, filtered, and concentrated. Nothing deposited from the lead subacetate fraction, but from the mercuric nitrate fraction needle-like crystals, interspersed with further crystals of asparagin, were found. The quantity was too small for identification, although the crystals answer the description of the glutamin identified by Zerban (*22*) in sugar-cane juice, and glutamin almost invariably accompanies asparagin in plant juices.

[1] These crystals were soluble in 5 per cent hydrochoric acid, insoluble in dilute ammonium hydroxid and dilute acetic acid.

To sum up the work on identification of compounds in sorghum juice the following list is given:

Sugars:
- Sucrose.
- Dextrose.
- Levulose.

Organic acids:
- Aconitic.
- Citric.
- Malic.
- Tartaric.
- Oxalic.

Polysaccharides:
- Starch.
- Galactans (in gums).
- Pentosans (in gums).
- Xylose (in cellulose of pith).

Nitrogenous compounds:
- Protein.
- l-Leucin.
- d-l-Asparagin.
- Glutamin.
- Cystin (?).
- Aspartic acid (?).

(3) DISTRIBUTION OF SUGARS.—From the practical standpoint, it is of importance to know the relative concentration of sugars in the various joints of the cane, since it may be unprofitable to mill the whole stalk; and from the standpoint of the physiology of the plant it is of interest to know how the concentration of sugars indicates the relative maturity of the various joints.

It is important to know also the sugar content of the leaves and of the suckers. It has been conclusively shown by many investigators (*5, p. 142; 19, p. 65*) that there is considerable sugar in the leaves but that the purity of the juice (percentage of the total solids as sugars) is so low that its sirup-making qualities are much inferior to those of the juice of the cane. Since most cane is milled either with the leaves removed or when the leaves are partially dried and hence contain but little extractable juice, the question of the leaf juice is of little importance and will not be dealt with further. The question of the juice of suckers, however, is of more importance, since under some conditions sorghum suckers badly; and when a corn binder is used for harvesting, the sucker canes are included. Here again many analyses are on record which show consistently that the suckers have a composition very similar to that of the main canes at the same stage of maturity. Since the suckers are always several stages behind the main canes in development, and since the maturity of a plot is judged by the seed heads of the main canes, the effect on the juice of cutting the two at the same

time is apparent. Collier (*5, p. 137*) says, "The suckering then of the crop, or at least the careful exclusion of suckers from that portion of the cane which is intended to be worked for sugar, is of the most imperative importance. For sugar production they are far worse than worthless. But they may be used for the manufacture of syrup, since both glucose and sucrose enter into its composition; and, in fact, the presence of the suckers in the crop would very easily prevent the crystallization of the syrup which the manufacturers of syrup frequently find a serious disadvantage." Since the analyses of suckers at this station contribute nothing new to the above facts, they will not be given here.

Collier (*5, p. 225–237*) determined the amount of sugars in the top and bottom halves of the cane and found little difference between them. In other experiments he divided the cane into thirds and again found little or no difference in the sugar content. So far as is known by the writers, no one has analyzed each joint separately. Reasoning by analogy to the suckers, the relative immaturity of the upper joints and the relative old age of the lower would lead one to expect a greater concentration of sucrose in the middle joints and of reducing sugars in the top and bottom joints.

The individual joints of several samples of cane in the dry dough stage were analyzed, with the results shown in figure 16. The concentration of sucrose and of reducing sugars varies inversely; but the variation is not proportional, since the total sugars are far higher in the middle portions of the cane. In fact, one of the most significant curves in the chart is that for the total sugars extractable per 1,000 parts of cane. This varies from 21 in the top joint to 46 in the middle and 25 in the bottom.[1] From the standpoint of sirup making, the top joint, and perhaps the bottom, could well be excluded from the milling, since they contain not only a small amount of sugar but a large amount of nonsugar solids (see the top curve in the graph), which are detrimental to good sirup making. Calculation shows that about 5 per cent of the total sugar would be lost by this practice, but this would be offset by the improvement in quality of the sirup. Went (*17*) finds about the same distribution of sugars in sugar cane as is reported above, except that there is a continuous increase in sucrose up to the joint next to the bottom.

In most plant juices the levulose exceeds the dextrose in amount, which fact is usually explained on the ground that the dextrose is more easily utilized in respiration. In sorghum juice the dextrose is always in excess of the levulose. A small portion of this dextrose may represent starch that is not yet polymerized (see next subsection for starch content of juices). It will be noticed that the excess of dextrose over levulose is least in the

[1] In explanation of the apparently very small amount of sugars extracted in the cane, it should be stated that this work was done with a small experimental mill which extracted an average of only 33 per cent of juice from the cane, whereas large mills obtain from 60 to 70 per cent.

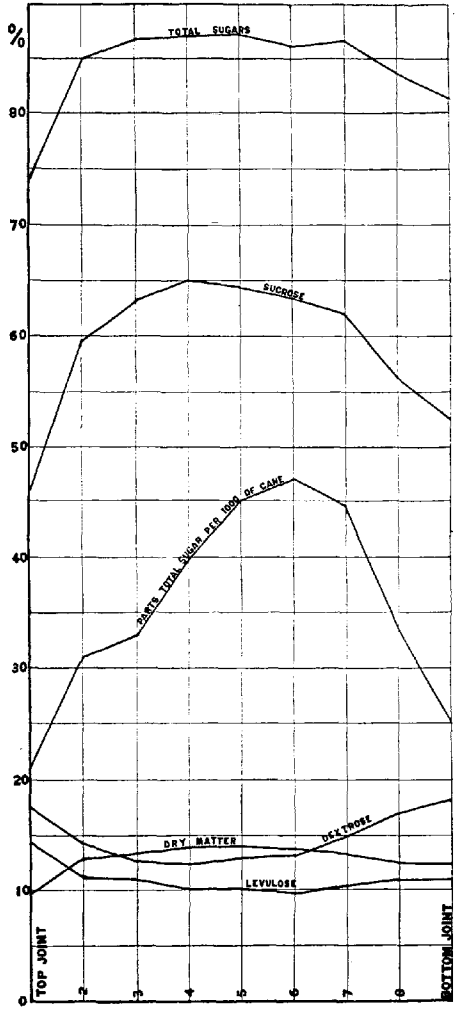

FIG. 16.—Distribution of the sugars and other solids in the various joints of sorghum cane.

upper portion of the cane, where respiration is proceeding most rapidly. This fact is in accordance with the view that the excess dextrose is the sugar most available for respiration. That portion of the dextrose which is equal to the levulose represents, of course, the invert sugar portion to be converted into sucrose.

(4) DEVELOPMENT OF THE CONSTITUENTS OF THE JUICE.—Many thousands of analyses have been made of the juice of sorghum to show the development of the various constitutents of the juice, especially the sugars. It would be fruitless to review them here, since in general they all show about the same trend of development—an increase of sucrose up to maturity, a decrease in reducing sugars, a concentration of the juice due to desiccation of the ripe cane, and a slight increase in purity (percentage of total solids as sugars).[1]

The results of the analyses at this station bring out very similar relations among the constituents of the juice. Since nothing new would be contributed by presenting all the analyses made at this station, only the work of the season of 1914 is given (fig. 17). This includes the separate determinations for levulose and dextrose, which have not been reported heretofore for sorghum juice during a whole season's development. For the sake of direct comparison, the only determinations for starch which were made, which were on the 1916 crop in connection with topping experiments, are also included in the graphs. Another fact that should be pointed out is that these analyses were made on cane grown at the northern limit of the sorghum-growing regions of the country and hence form a basis of comparison with cane grown during a longer and warmer season. To make this comparison more apparent, Collier's curves (*5, p. 169*) for the average results of analyses of Early Amber cane grown at Washington, D. C., in 1881, are also given in figure 18.

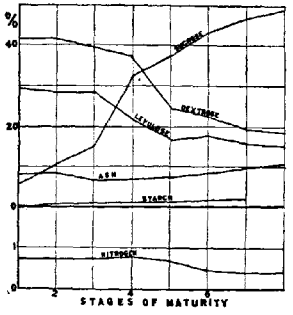

FIG. 17.—Development of the constituents of the juice of sorghum. (See p. 2 for description of the stages of maturity.)

One or two significant facts can be pointed out in the curves in figure 17. In the first place, the ratio of levulose to dextrose changes during the progress of the development of the plant. As maturity approaches, the dextrose decreases more rapidly than the levulose. At the same time, the starch increases from about 0.7 per cent to 1.8 per cent. This small amount of starch can not account for the marked decrease in dextrose.

[1] It should be pointed out that in sorghum the standard for purity is the total sugars, whereas in sugar-cane and sugar-beet work it is the sucrose alone. This is because of the fact that in sirup making the total sugars are utilized.

The latter is probably due to at least three causes: (1) Respiration, (2) conversion into sucrose, and (3) conversion to a slight extent into starch. The changes in the dextrose-levulose ratio in the juice during growth and in the various joints of the cane (fig. 16) are in the reverse order. Thus, from the standpoint of maturity, the lower joints contain the most dextrose while the older plants contain the least. No explanation is offered for this apparent anomaly.

Another significant fact brought out by the curves is the undiminished upward trend of the sucrose curve clear to the last stage analyzed. In this stage the plant was mature, judged by the condition of the seeds,

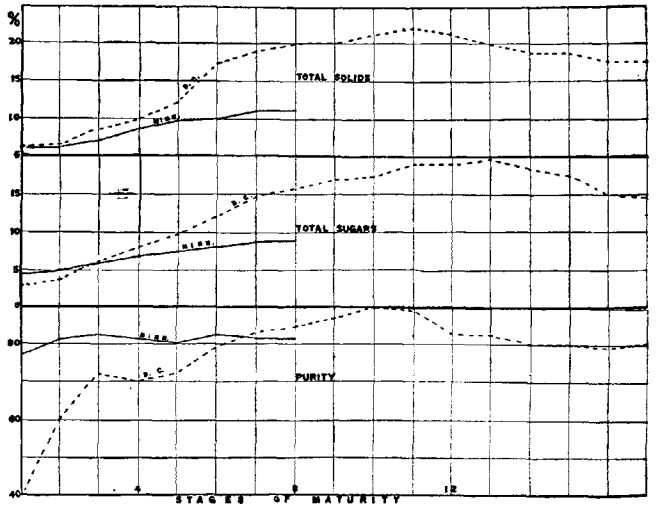

FIG. 18.—Comparison of Early Amber sorghum grown in Minnesota with that grown in the District of Columbia.

since the latter were hard and starchy and showed fair germination. It is apparent, however, that the changes in composition of the juice had not ceased. If the taking of later samples had not been prevented by frost, there is no doubt that the sucrose content would have undergone still further increases. In figure 18 there is clearly shown a continuation of increases in total sugars, dry matter, and purity in Collier's Virginia-grown cane during a period of 10 to 20 days after apparent maturity. Sorghum cane grown as far north as Minnesota probably never reaches the maximum possible content of sugars. The data in the curves are for 1914, which was a dry and hot season during the period of maturation of sorghum. Although this plant can withstand these climatic conditions

better than most crop plants, it can not develop normally, as is shown by the low content of solids in these samples and by the high purity of of the juice during the early periods. As regards the latter, it should be kept in mind that the "purity" means the relative proportion of total sugars in total solids. In an immature cane the purity is low, as is shown by the curve for the Virginia samples. The relatively high purity of the Minnesota samples is another example of the oft-observed fact that during periods of unfavorable growth conditions a plant attempts to reach maturity as quickly as possible. Usually this is evidenced in the reproductive parts alone; in the sorghum it is apparent also in the composition of the juice.

The curve representing the percentage of nitrogen in the juice remains practically level. This signifies that the absolute amount of nitrogen must increase clear to maturity. The nitrogen compounds comprise a prominent portion of the nonsugar solids, and the flatness of the nitrogen curve is in harmony with that of the purity curve.

VI.—EFFECT OF REMOVING THE SEED HEADS

There are many references in the literature on sugar-producing plants relating to the effect on the composition of the juice of removing the fruiting parts of the plant. Collier (*p. 138–140*) found that removing the seed heads at immature stages hastened the maximum production of sugar in the juice, but that this maximum was the same as that in unheaded stalks, although in the latter it was reached a week or 10 days later. Thus the effect of heading the cane was to hasten the maturity of the juice but not to increase its potential sugar content. Wiley (*19*) reports analyses which show very slight differences in favor of topping. Heckel (*9*) reports an increase in sucrose in both corn and sorghum due to removal of the floral parts. It is not clear from the latter data, however, whether only the sucrose is increased or the total sugars as well. Other work shows similar inconclusive data on this question.

For two seasons heading experiments were conducted at this station. The results are presented in Tables II and III. It will be seen that in general the data corroborate Collier's conclusions that heading merely hastens the maturity of the juice but does not affect its final composition. There is some evidence that the amount of starch produced is increased, but the differences are too small and the number of analyses too limited to warrant any definite conclusions. From the standpoint of sirup manufacture, the removal of the seed heads should be practiced in order to bring about an earlier maturing of a portion of the crop, although by so doing the value of the seed would be lost.

TABLE II.—*Effect of the removal of the seed heads on the composition of the juice*

1915 CROP

Treatment of plants.	Stage of growth when heads were removed.	Stage of growth when analyses were made.	Percentage of total solids.	Percentage of total solids as—	
				Sucrose.	Reducing sugars.
Heads off	Full bloom	Hard dough	18.0	71.7	10.1
Heads on	do	do	14.9	65.3	18.5
Heads off	Milk	Soft dough	13.0	56.9	30.0
Heads on	do	do	11.2	47.4	37.0
Heads off	Milk	Hard dough	14.3	60.8	27.5
Heads on	do	do	10.5	50.6	35.6
Heads off	Milk	Mature	15.8	64.9	18.0
Heads on	do	do	12.9	56.7	24.4
Heads off	Soft dough	Hard dough	11.6	50.9	33.0
Heads on	do	do	11.4	57.0	27.2
Heads off	Soft dough	Mature	12.8	62.8	22.0
Heads on	do	do	11.2	52.5	30.6

TABLE III.—*Effect of the removal of the seed heads on the composition of the juice*

1916 CROP

Date on which heads were removed.	Stage of growth when heads were removed.	Stage of growth when analyses were made.	Percentage of total solids.	Percentage of total solids as—		
				Sucrose.	Reducing sugars.	Starch.
Aug. 24	Panicles out	Panicles out	7.1	15.4	68.9	0.7
Sept. 7	do	Milk	13.1	41.0	38.8	2.6
18	do	Soft dough	13.6	44.3	34.0	2.1
26	do	Hard dough	14.5	63.4	25.1	.....
Aug. 24	do	Panicles out	7.1	15.4	68.9	.7
Sept. 7	do	Milk	12.4	48.3	30.9	2.8
18	do	Soft dough	13.7	42.9	39.7	1.6
26	do	Hard dough	10.7	43.8	42.7	.....
Aug. 24	Full bloom	Full bloom	7.7	27.1	58.5	1.0
30	do	Milk	10.4	36.3	45.0	1.5
Sept. 8	do	Soft dough	15.8	49.8	27.2	3.1
18	do	Hard dough	15.2	49.8	33.2	2.6
26	do	Mature	15.9	64.7	23.2	.....
Aug. 30	Milk	Milk	10.8	39.6	42.3	1.2
Sept. 7	do	Soft dough	14.2	58.5	24.4	2.8
18	do	Hard dough	14.5	63.3	23.2	2.7
26	do	Mature	17.0	41.1	41.4	.....
8	Soft dough	Soft dough	12.8	43.5	31.2	1.7
18	do	Hard dough	13.1	60.4	24.3	1.6
26	do	Mature	13.9	43.0	40.7	.....

VII.—SUMMARY

(1) Three varieties of sorghum cane were used for studying the progressive development of the plant and the chemical composition of the various parts of it.

(2) The relative proportion of leaves, seed heads, and clean cane by weight in both the fresh and the partially dried condition was determined for one season.

(3) Considering the whole plant, there was found to be a continual increase in dry matter up to maturity. The percentage of crude fiber decreases at practically the same rate as that at which the soluble carbohydrates increase. The crude fat, ash, and protein percentages remain almost constant throughout the periods of growth studied.

(4) The computation of the total quantities of each constituent present in the plant at the various stages of growth brings out the fact that this plant builds up during the earlier part of the season its cellular structure of fiber, protein, and mineral matter, and that the later stages of growth consist in the filling up of these tissues with carbohydrates (starch in the seed, sugars in the stalk).

(5) No evidence was found which would indicate that the leaves are deprived of their carbohydrates to supply the stalk, at least during the periods of growth studied. The older the plant, the higher is the feeding value of the leaves.

(6) The maturation of the seed heads consists almost entirely in the filling out of a fiber and protein framework with starch.

(7) There is a considerable accumulation of mineral matter in the leaves, due probably mostly to calcium and silicon.

(8) Large quantities of juice were employed in isolating and identifying the nonsugar solids. Equal volumes of alcohol threw down a preprecipitate which consisted of three portions: (a) proteins, (b) cellular material in suspension, arising from the crushing of the fiber in the mills, and (c) true gums.

(9) The gums are complexes of galactan and pentosans, with about 20 per cent of mineral matter, principally calcium, magnesium, and potassium.

(10) The organic acids found in sorghum juice are aconitic, malic, citric, tartaric, and oxalic.

(11) Nonprotein (amid) nitrogen is very high in sorghum juice, even in mature cane. This is an important contributing factor in the difficulties of defecating sorghum juice for either sirup or sugar production.

(12) The following nitrogenous substances were identified in sorghum juice: l-leucin, d-l-asparagin, glutamin, cystin (?), and aspartic acid(?).

(13) The juice of suckers has a composition similar to that of the main canes at the same stage of maturity. They are, however, usually from one to three weeks behind the main canes in maturity.

(14) The middle joints of the cane are higher in total sugars and in sucrose but lower in dextrose and in levulose, than the upper and the lower joints. The upper joint contains so little sugar and such a low coefficient of purity that it can well be excluded from the milling in sirup making.

(15) Sorghum cane grown in Minnesota has a much lower sugar content than cane grown in regions of longer and warmer growing seasons. There are indications that if the advent of frost could be delayed, cane which is usually considered mature would continue for another week or 10 days not only to increase the ratio of sucrose to reducing sugars but to elaborate more total sugars. The juice of northern-grown cane has a higher purity than that of southern grown. This is a phenomenon of early maturation exhibited by most plants when grown under suboptimum conditions.

(16) At the time of the first appearance of the panicles the reducing sugars are greatly in excess of the sucrose. The former rapidly decrease and the latter rapidly increases, until at the stage of full bloom they are about equal in amount. The respective changes continue up to maturity, when the ratio of sucrose to reducing sugars is in Minnesota-grown cane about 70 to 30. In very mature Virginia-grown cane the ratio is 90 to 10, or even higher.

(17) Removal of the seed heads prior to maturity hastens the production of the maximum amount of sugar in the juice, but the same maximum would be attained later without the removal of the seed heads.

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SIMPLE METHOD FOR MEASURING THE ACIDITY OF CEREAL PRODUCTS: ITS APPLICATION TO SULPHURED AND UNSULPHURED OATS[1]

By VICTOR BIRCKNER

Mycologist in Fermentation Investigations, Bureau of Chemistry, United States Department of Agriculture

INTRODUCTION

Much attention has been given during recent years to acidity determinations in connection with the analysis and valuation of cereals and their various products. Some of the newer literature on this subject is reviewed in a recent article by Lüers and Adler (*11*).[2] Of the methods proposed by various investigators, that of Schindler (*12*) has been rather extensively used in this country (*5, 8, 18*), especially in work with corn (*Zea mays*). Its use is also recommended in a recent French paper by Leprince and Lecoq (*9*).

Besley and Baston (*5*) alone have investigated approximately 10,000 samples of corn by means of a modification of Schindler's method; and the acidity values thus obtained are considered by Winton and his coworkers "the best chemical means of detecting actual spoilage, or at least a tendency in that direction" (*18, p. 1*). The method involves extraction of the material for 16 to 18 hours at room temperature with neutral alcohol (specific gravity 0.86) and titration of the alcoholic extract with standard alkali. The acidity is stated in terms of cubic centimeters of normal alkali required to neutralize the acidity of the extract from 1,000 gm. of material. The acidity figure 30 is taken as an arbitrary limit, beyond which the value should not rise for sound corn.

This alcohol extraction method has several weak points, as will at once become apparent if we consider for a moment the nature and mode of formation of the acids present in grain extracts. Lüers and Adler (*11*) have shown conclusively that in extracting barley or malt, acid-forming ferments come into play, and that it is therefore necessary to make a distinction between the original acidity of the material and the acidity formed during extraction. According to the same authors, the acidity of barley or malt extractions is due mostly to acid phosphates, which are partly present as such and partly are formed during extraction from organic phosphorus complexes through the action of specific ferments, to which they give the name phosphatases. The

[1] An abstract of this paper was read before the thirty-third annual convention of the Association of Official Agricultural Chemists at Washington, D. C., in November, 1916.

[2] Reference is made by number (italic) to "Literature cited," p. ——.

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solubility of those acid phosphates is declared to be greatly diminished in the presence of alcohol, a fact which had previously been recognized by Weiss (*17*) and by Swanson (*14*, *15*). Weiss found that if ground barley is first extracted with 96 per cent alcohol a subsequent extraction with water yields a far smaller quantity of soluble phosphorus compounds than a direct water extraction. Of the total phosphoric acid in 100 gm. of barley, 79 per cent was found to be normally soluble in water by 18 hours' extraction at room temperature with addition of toluol. If the same material was first treated with 96 per cent alcohol in a Soxhlet extractor for 20 hours, a subsequent extraction with water yielded only 45 per cent of the total phosphoric acid contained in the meal, while not more than 3.66 per cent had been removed by the alcohol. Weiss seeks to explain this difference by assuming that the high phosphorus content of the water extract is due to the action of enzyms, which he thinks are destroyed by the alcohol treatment. This explanation, however, is not in harmony with the more recent findings of Adler (*1*, *2*, *3*). This investigator has made extensive studies of the various groups of acid-producing ferments which come into play when ground barley or malt is extracted with water. He found that these ferments, and particularly the phosphatases, are very resistant not only to the action of the common disinfectants such as chloroform, toluol, hydrogen peroxid, etc., but also to dry heat, and that their activity is not impeded by cold alcohol, even if present in high concentrations.

It does, of course, not necessarily follow that the results obtained with barley by these investigators will apply, without qualification, to corn, oats, or rye. However, there is a strong probability that similar conditions will prevail in all gramineous seeds; at any rate, the results obtained with barley in Germany deserve the full attention of everyone who is working along similar lines with other cereals. Schindler, for example, would probably have hesitated in devising his above-mentioned alcohol extraction method for determining corn acidity had he read the paper of Weiss (*17*) which was published two years previously.

While recognizing the impossibility of suppressing acid-producing ferments by means of cold alcohol, Lüers and Adler (*11*) found that a brief boiling with 96 per cent alcohol completely destroys these ferments. They therefore make this boiling with neutral 96 per cent alcohol part of their method for measuring the original acidity of ground barley and malt. During the boiling, which is done in open beakers with occasional stirring, most of the alcohol is driven off, and when the material has reached a doughy consistency it is allowed to cool. Distilled water is then added and the mixture permitted to stand for several hours with occasional stirrings and the addition of toluol. The weight of the total amount of liquid is then determined, and after the liquid is filtered a suitable aliquot is titrated with *N*/10 alkali. Lüers and Adler are well aware of

certain inaccuracies of this method. For example, they point out that by boiling the grain material with 96 per cent alcohol certain proteins are coagulated and that in being thus transformed these bodies possess the power either to absorb acids or to combine with them. Part of the grain acidity might thus be occluded and escape detection. However, these authors recommend the above procedure for want of a better means of measuring the quantities of acid originally present in barley or malt.

ACIDITY VALUES OBTAINED BY THE ICE-WATER METHOD AS COMPARED WITH THOSE OBTAINED BY THE SCHINDLER METHOD

The method proposed in this paper was first used during a study on the diastatic power of oats. In that study the ice-water extraction method recommended by Thatcher and Koch (*16*) was employed; but it was thought necessary to neutralize each oat extract before determining its diastatic strength, inasmuch as the oats in question represented different degrees of soundness and some of them had been subjected to the sulphuring process as now practiced by the trade (*13*). In neutralizing the different aqueous oat extracts with *N*/10 sodium hydroxid very pronounced differences in acidity were observed in samples which, when the above-mentioned aclohol extraction method of Schindler was used, had shown no appreciable difference in acidity. Furthermore, the values obtained by simple titration of the ice-water extracts appeared to represent the actual acidity of the respective sample far more truly than those obtained by means of the Schindler method. The data compiled in the subjoined table will serve to illustrate these relations. The second and third columns of figures denote cubic centimeters of normal alkali which were required to neutralize the acidity in the extract from 1,000 gm. of dry material.

The samples 1s to 10s in the following table represent the same oats as No. 1 to 10. The latter set had been taken before, and the former after, the sulphur bleaching. These duplicate samples of oats of the 1915 crop had been secured by official inspectors at commercial elevators in Chicago. Since most of these oat shipments when arriving at the Chicago market appeared discolored or otherwise damaged, the dealers had tried to improve their appearance by means of sulphuring.

TABLE I.—*Acidity values of freshly ground oats and of corn meals, shown by two different methods*[1]

Sample No.	Schindler's alcohol extraction method.	Ice-water extraction method.
Unsulphured oats:		
1	33.6	18.3
2	48.2	18.5
3	41.2	17.3
4	41.0	13.4
5	45.0	21.7
6	36.5	17.0
7	42.4	17.0
8	49.2	16.3
9	42.0	11.3
10	44.0	14.9
Sulphured oats:		
1S	55.0	91.8
2S	34.6	54.0
3S	46.8	46.9
4S	47.0	42.8
5S	41.6	41.5
6S	35.6	35.6
7S	28.2	28.4
8S	53.3	27.5
9S	42.6	20.0
10S	39.0	27.4
Corn meal (degerminated):		
11	13.0	11.0
12	31.6	10.1
Corn meal (whole kernel):		
13	109.6	106.0
14	154.4	59.0

[1] Expressed in cubic centimeters of normal alkali required to neutralize the extract from 1,000 gm. of material.

In making the above determinations, all samples, after being ground to the same degree of fineness, were dried over calcium chlorid in a large air-tight cabinet with an inside temperature of 38° to 40° C., the air being kept in circulation by means of an electric fan. This mode of drying at low temperature was adopted because the oat samples, as indicated above, were also tested afterwards for their diastatic strength. The ice-water extractions were carried out in a cold-storage compartment of the Bureau of Chemistry, which is kept at a temperature close to 1° C. Distilled water of the same temperature was kept on hand. A weighed quantity of dried sample, usually 16 gm., was placed in the proper extraction bottle and the latter allowed to stand in the cold-storage room until its contents had reached the desired temperature. One hundred and sixty cc. of precooled distilled water were then pipetted into each bottle, the latter closed with a tightly fitting rubber stopper and shaken vigorously at 15-minute intervals. At the expiration of the extraction period (one hour for oats) the contents of each bottle were poured upon dry folded filters (S. & S.

No. 588 Falten filter). By pouring back the first few cubic centimeters of filtrate a homogeneous extract was obtained which, as a rule, was clear and easy to filter when it was obtained from sulphured oats. The extracts from unsulphured oats filtered more slowly and gave turbid filtrates. This turbidity, however, did not interfere with the subsequent titration. Usually when the ice-water extracts of sulphured oats were neutralized a white amorphous precipitate was formed, while with unsulphured oats no such precipitation took place. With the corn meal samples 13 and 14 similar precipitations occurred upon neutralizing. It was thought that through the high acidity of these samples some constituents had been dissolved which are insoluble in neutral liquids. However, this explanation does not cover all cases, since precipitation also occurred upon neutralizing the ice-water extracts of sample 7s, 9s, and 10s in the foregoing table, as well as the extracts of unsulphured samples 63 and 74 in Table V. Since the latter samples were two years old it is possible that the age of a sample bears some relationship to the formation of this neutralization precipitate. All manipulations of the ice-water method were carried out in the cold-storage compartment with the exception of the titrations. For the latter, a suitable aliquot of the respective filtrate was pipetted out, usually 40 cc. Phenolphthalein was very satisfactory as indicator, giving a sharp endpoint. It was employed in a 0.5 per cent solution in 50 per cent alcohol, not more than five drops were added to any of the aliquots, and the appearance of a faintly reddish tint was taken as the endpoint of titration. Methyl red could not be used in this work. In determining the acidity by the alcohol extraction method the directions given by Besley and Baston (5) were followed. The endpoint of titration of the alcoholic extract is not very sharp, a fact which was recognized by Besley and Baston in their bulletin and which constitutes one of the drawbacks of their method.

From Table I it is seen that the acidity values of natural unsulphured oats obtained at commercial elevators are in every case higher if determined by Schindler's alcohol method than if determined by the simple ice-water extraction method. If the latter values are used as a basis of calculation, these differences show an average for samples 1 to 10 of well over 250 per cent, the smallest difference being 184 per cent and the largest 372 per cent. They are therefore quite appreciable and are doubtless to a large extent attributable to the alcohol content (7) of the extract made by the Schindler method. Differences of the same general kind are shown by the corn meal samples 11 to 14, all of which had been milled three years previously, samples 12 and 14 having been dried with artificial heat and samples 11 and 13 having been stored without drying.

After being bleached with sulphurous acid, oats would naturally be expected to possess a higher degree of acidity than before the bleaching.

As shown in Table I, this view was confirmed in every instance where the acidity had been determined by the ice-water method. For example, with sample 1 the acidity before bleaching was 18.3, and after bleaching 91.8, or five times as high. While less pronounced in samples 2 to 10, the rise in acidity due to sulphuring was distinctly evident in each case. With the Schindler method, as modified by Besley and Baston, the increase in acidity due to sulphuring was hardly, if at all, apparent. In fact, in 5 of the 10 cases the figure for the sulphured sample was even lower than that for the same oats before sulphuring.

When comparing the results obtained by the two methods on the sulphured samples 1s, 2s, 3s, and 7s (see Table I), we find that, contrary to the results with unsulphured samples, the alcohol extraction method gave here lower values than the ice-water method. In other words, while practically all the acid of the oat had passed into the ice-water extract in an hour's time, the 18-hour digestion with 80 per cent alcohol had not effected the solution of all acid constituents. Thus, for sample 1s the ice-water method yielded the acidity figure 91.8; and Besley and Baston's modification of the Schindler method yielded only the figure 55, a deficiency of 40 per cent. While it would not be difficult to offer an explanation for this finding [1] I will confine myself here to a comparison of the values obtained by the two methods.

From the above experimental results one can not escape the impression that there are a number of objections to the use in work of this kind of the Schindler method as modified by Besley and Baston. Briefly summarized, these objections are:

(1) The lack of a definite endpoint of titration.

(2) The misleading character of the results obtained by the method, inasmuch as:

(*a*) The acidity values of raw, unsulphured cereals are invariably too high.

(*b*) The acidity values for sulphured cereals (oats) are frequently too low.

(*c*) The increase in acidity which always occurs when grain is sulphured frequently fails to find expression in the results, the latter quite often indicating instead of the actual increase an apparent decrease in acidity upon sulphuring, due to incomplete extraction of the acid.

The above statements refer to results obtained with freshly ground material. It may be mentioned also that for the grain dealer or manufacturer, as well as for the commercial chemist, the high price of pure alcohol would make this method rather expensive. The cost factor would, of course, be of only secondary importance, provided that the

[1] I may only point to the close similarity between this result and the findings of L. Weiss (17), which are discussed in the introduction of this paper.

method furnished as accurate a means for measuring grain acidity as has been claimed by its users. Unfortunately, my findings in the above Table I in conjunction with those recorded in the previous paper (7) point to the contrary conclusion.[1]

While for work on freshly ground sulphured or unsulphured oats the Schindler method is clearly inadequate, yet the figures obtained for corn by Besley and Baston (6) would seem to bear a certain relation to the relative age or soundness of the respective samples. But the changes which they have measured can not represent true acidity only. They are doubtless due, in a large measure, to the formation of amphoteric protein cleavage products which cause the titration value to increase suddenly in the presence of alcohol to several times its former magnitude.[2]

It was thought advisable that another chemist should examine a number of cereal products for acidity by both the proposed ice-water method and the Schindler method as modified by Besley and Baston. Mr. M. G. Mastin of the Bureau of Chemistry kindly undertook this task, and his findings are given in Table II.

[1] In the latest modification of their method Besley and Baston (6) apply an electric mixing apparatus by which they find it possible to reduce the time of extraction in their alcohol method to 30 minutes, as against 16 to 18 hours, the time prescribed in their former paper. The introduction of this electrical device, which by the way must be rather expensive, is merely an improvement in the technic. Had the authors at the same time abolished the use of alcohol, a real improvement of their method would have been attained. Since in the 18-hour extraction with alcohol the latter had been prescribed for the purpose of preventing bacterial or enzymic action, there seems to be no longer any reason for its use, if the time of extraction is reduced to 30 minutes. From the data given in this and the preceding article, it is evident that the errors introduced by the use of alcohol must far outweigh its possible usefulness in a 30-minute extraction.

[2] One might be tempted to believe that, since by using Besley and Baston's method the acidity values of freshly ground, untreated cereals appear to be always higher than the values obtained by extraction with ice water, it should be possible to calculate the ice-water acidity from the figures published in Besley and Baston's paper (6). However, this would not be permissible, since the higher values obtained by their method depend not alone on the concentration of the alcohol, which is the same in each case, but also on the nature and concentration of those amphoteric grain constituents which are responsible for the shift in acidity in the presence of alcohol. The concentration of these bodies naturally varies from case to case.

TABLE II.—*Further comparisons between the ice-water and the alcohol method, showing increase in acidity which takes place in ground cereals upon standing*[1]

[Determinations made by Mr. M. G. Mastin]

Sample No.	Crop year.	Schindler's alcohol extraction method.	Ice-water extraction method.	Remarks.
Unsulphured oats:				
15	1916	42.9	16.7	Freshly ground.
16	1915	27.5	19.8	Do.
17	1916	39.0	13.7	Do.
18	1915	30.0	13.8	Do.
18r	1915	.....	31.0	After standing.
19	1916	39.0	16.7	Freshly ground.
20	1915	32.0	17.0	Do.
21	1915	27.2	14.0	Do.
21r	1915	104.0	24.0	After standing.
22	1915	15.4	14.5	Freshly ground.
22r	1915	129.0	37.5	After standing.
23	1914	13.4	8.5	Freshly ground.
23r	1914	57.0	20.2	After standing.
23rr	1914	149.0	43.0	After long standing, moldy.
24	1914	13.6	11.5	Freshly ground.
24r	1914	82.2	21.5	After standing.
25	1914	13.6	13.0	Freshly ground.
25r	1914	56.6	22.5	After standing.
26	1914	20.6	15.0	Freshly ground.
26r	1914	121.0	46.0	After standing, moldy.
27r	1914	88.8	35.0	After standing.
27rr	1914	173.5	38.6	After long standing, moldy.
28r	1914	136.6	36.0	After standing.
29r	1914	47.6	19.0	Do.
30r	1915	63.0	20.0	Do.
31	1916	36.0	22.0	Freshly ground.
32	1916	26.6	12.0	Do.
33	1916	26.6	16.0	Do.
34	1916	36.6	17.0	Do.
35	1916	35.9	18.0	Do.
36	1916	36.0	16.0	Do.
37	1916	41.5	16.5	Do.
38	1916	53.7	17.3	Do.
39	1916	60.5	16.0	Do.
40	1916	52.5	14.7	Do.
41	1916	26.0	16.0	Do.
42	1915	34.2	19.3	Do.
Sulphured oats:				
15s	1916	48.7	21.0	Do.
16s	1915	37.2	40.3	Do.
17s	1916	35.0	32.7	Do.
18s	1915	28.0	22.5	Do.
18sr	1015	126.0	21.6	After standing.
19s	1916	29.0	33.0	
20s	1915	28.6	34.5	Freshly ground.
21s	1915	67.8	78.0	Do.
21sr	1915	78.4	77.7	After standing.

[1] Expressed in cubic centimeters of normal alkali required to neutralize the extract from 1,000 gm. of material.

TABLE II.—*Further comparisons between the ice-water and the alcohol method, showing increase in acidity which takes place in ground cereals upon standing*—Continued

Sample No.	Crop year.	Schindler's alcohol extraction method.	Ice-water extraction method.	Remarks.
Sulphured oats—Contd.				
43	1915	33.8	36.5	Freshly ground.
44	1916	29.8	15.0	Do.
45	1916	35.3	31.7	Do.
46	1916	33.4	36.0	Do.
47	1915	45.6	18.3	Do.
48r	1915	97.4	31.1	After standing.
49	1915	37.5	39.5	Freshly ground.
50	1915	40.0	22.7	Do.
Unsulphured barley:				
51	?	14.8	10.0	Do.
52	?	24.8	22.0	Do.
Sulphured barley:				
51s	?	21.5	18.0	Do.
52s	?	21.4	32.0	Do.
Yellow corn meal (whole kernel):				
53	?	26.4	69.5 (27.8)	No rancid odor.
54	?	15.8	33.0 (23.4)	Do.
55	?	20.6	35.0 (19.0)	Do.
White corn meal (degerminated):				
56	?	48.2	21.5 (22.0)	Do.
57	?	46.4	22.0 (21.8)	Do.

While in the work recorded in Table I all oat samples had been freshly ground before being extracted, a number of the samples listed in Table II had been ground about eight months before being handed to Mr. Mastin for analysis. These old samples gave off a more or less rancid odor, and some of them had become infested with molds. Samples of this type are marked in the table by adding the letter "r" to the original number and also by the descriptive remark in the last column. In some cases the figures obtained for the same oats when freshly ground are also given. In general, the values connected by brackets refer to the same sample of oats. Thus for sample 23 the ice-water value 8.5 was obtained for the freshly ground oat; the corresponding figure, after the sample had been ground and kept in a glass jar for several months, was 20.2; and for another sample which was moldy and more strongly deteriorated the value was 43.0. In the series of sulphured samples those marked with the letter "s" correspond to samples with the same number in the unsulphured set. In other words, a sample of a given lot of oats or barley was taken both before and after sulphuring; so the values listed under 17 are the values for the oat of that number before sulphuring, and those under 17s are the values for the same oat after it had been sulphur bleached.

Samples of the sulphured oats No. 18s and 21s which had been kept for a long time after grinding were also available for investigation,

and the acidity values found are listed under No. 18sr and 21sr, respectively. It is noticeable that these sulphured samples showed no increase in the ice-water acidity even though they had stood for a prolonged period after being ground. It would seem, therefore, that the increased titration values found in these instances with the Schindler method are wholly due to the formation, on standing, of amphoteric protein deterioration products. The latter are not determined by the ice-water method, since by its use only substances are measurable which react acid in aqueous solution. In other words, the interesting fact is apparent from these results that with ground samples which prior to the grinding have been sulphur bleached the ice-water acidity does not increase on standing, while unsulphured oats with the same method and under like conditions always show an increase in acidity. The sulphur bleaching, therefore, has probably destroyed the acid-forming ferments of the grain. It has not, however, stopped the decomposition of the proteins, and the rate of this decomposition is doubtless registered by the increased titration values obtained for these samples when Schindler's method is used. The latter method might, therefore, under certain conditions and when used in conjunction with a true acidity method, be developed into a useful measure for the rate of decomposition of proteins, as already pointed out in my previous paper.

The barley samples and many of the oats samples listed in Table II had been collected by me at various grain elevators in the Central West during the fall of 1916. The five samples of commercial corn meal No. 53 to 57 had been examined by me seven months previously by means of the ice-water method. The values then obtained are recorded in parenthesis for comparison. These corn meal samples were extracted with ice water for 1½ hours, as against 1 hour for barley and oats. The figures in Table II represent cubic centimeters of normal alkali which were required to neutralize the extract from 1,000 gm. of the material. The moisture content of the latter was not taken into account. The determinations were made in the laboratory, using an ice-water bath, in the manner described under the next heading.

The values obtained by the two methods in the extraction of the corn meals 53 to 57, which had been purchased in the open market, are also of interest.[1] The ice-water values of Mr. Mastin, compared with those which I had obtained seven months previously (the latter being shown in parenthesis) indicate a distinct rise in acidity for the whole kernel meals 53 to 55 but no rise for the degerminated meals 56 and 57. Possibly the latter meals had received some kind of chemical treatment which prevented the increase in acidity in a similar manner as the sulphur bleaching had prevented it in the oat samples 18sr and 21sr. Another explanation would be that the acid-forming ferments are all located in the germs and had been removed with the latter. Unfortunately I had

[1] The yellow corn meals 53 to 55 had been reground to about the same fineness as samples 56 and 57.

employed only the ice-water method, and not the alcohol method, when first examining these corn meals. From the figures obtained by Mr. Mastin it is also clear that by extracting samples 53 to 55 with alcohol in the manner prescribed by Besley and Baston, it was not possible to bring into solution all of the acid-reacting constituents of these meals.

FURTHER STUDY OF THE ICE-WATER EXTRACTION METHOD

The following experiments were carried out by me for the purpose of studying more closely the conditions and limitations of the ice-water extraction method.

It was thought that a brief boiling might possibly give some indication of the character of the free acids present in ice-water extractions of oats and corn. Equal aliquots of filtered ice-water extracts were therefore pipetted into Erlenmeyer flasks and the one sample heated to boiling on an asbestos screen, while the other was titrated directly. The boiling was continued for 30 seconds, whereupon the flask was removed from the screen, cooled under the tap, and at once titrated.

TABLE III.—*Effect of heating upon the acidity of ice-water extractions of oats and corn*[1]

Sample No.	Titrated directly.	Titrated after boiling ½ minute.
Unsulphured oats:		
58	20. 0	19. 3
59	21. 0	20. 0
Sulphured oats:		
58s	50. 3	50. 7
59s	27. 3	26. 0
Corn meals:		
60	55. 0	55. 7
61	30. 7	29. 3

[1] Expressed in cubic centimeters of normal alkali required to neutralize the extract from 1,000 gm. of material.

It is seen from the foregoing table that the short heating had no appreciable influence on the titration values of these samples. The boiled liquids, moreover, showed no precipitate or other visible changes. Upon standing in the cold-storage room over night both the boiled and the unboiled extracts had turned acid again. It may be mentioned that I have regularly observed that ice-water extractions of oats or corn when filtered and kept at a temperature of 1° to 2° C. for over a day without being neutralized show no change in acidity. However, if such extracts are neutralized they form fresh acid upon standing and continue to do so after renewed neutralization. The presence of chloroform does not affect this acid formation. Apparently, therefore, the acidity found in each of these extracts represents a chemical equilibrium, which tends to become reestablished if disturbed by the process of neutralization.

The acidity figures in Table IV will serve to illustrate these relations. They are also intended to show the influence of the time of extraction upon the acidities of the extracts of corn and oats. Three portions of 12 gm. each of finely ground oats were extracted with 100 cc. of ice water, the bottles being shaken at 15-minute intervals. Three 12-gm. portions of a commercial corn meal were extracted in the same manner. All six extractions were started at the same time. At the end of 1 hour the contents of one bottle each of the oat and corn infusions were poured upon dry folded filters, and two 25-cc. aliquots of the filtrate pipetted into Erlenmeyer flasks. The one aliquot was titrated at once with *N*/10 sodium hydroxid, while the other aliquot was left in the cold-storage room for 24 hours before being titrated. The former aliquot, after being neutralized, was returned to the cold storage. At the end of 1½ and 2 hours, respectively, the filtration of another infusion of both corn and oats was begun, and the respective aliquots titrated in the manner above described. Together with the titration of those aliquots which had stood for 24 hours before being neutralized the aliquots neutralized on the previous day were retitrated. The alkali thus required was due to the fresh acid which had been formed after the extracts were neutralized These increments are listed in separate columns of the following table.

TABLE IV.—*Relation between time of extraction and acidity of ice-water extracts of oats and corn, and the formation of fresh acid after neutralization of the extracts* [1]

Time of extraction (in minutes).	Treatment of extract.	Oats.		Corn meal.	
		First titration.	Second titration.	First titration.	Second titration.
60	Titrated at once	0.66	0.21	0.73	0.24
	Titrated after 24 hours	.65	.....	.73	.....
90	Titrated at once	.69	.22	.86	.23
	Titrated after 24 hours	.68	.....	.85	.....
120	Titrated at once	.71	.21	.88	.21
	Titrated after 24 hours	.72	.....	.88	.....

[1] Expressed in cubic centimeters of *N*/10 sodium hydroxid required to neutralize 25 cc. of extract (3 gm. of material).

It is seen from the foregoing table that the amount of acid found in the extracts varies with the time of extraction. However, with oats only slight increases in acidity were observed if the extraction period was extended beyond 1 hour. With corn, the difference between the values for the 60- and 90-minute extractions was rather marked, whereas the difference between the 90- and 120-minute extractions was insignificant.

There are two possible explanations for the slight differences in the quantity of acids found in the three oat extracts of Table IV. One would be that the acids present in the material are dissolved very slowly

and the other that the total original acidity of this oat is included in the value found for the 60-minute extraction and that the rise upon prolonged extraction is due to ferment action. From my present experience I would prefer the latter explanation. While, as shown above, no acid-producing ferments come into play in the filtered, unneutralized grain extracts at the cold-storage temperature, it is quite conceivable that if the liquid remains in prolonged contact with the grain solids some insoluble constituents may be hydrolized by specific ferments and form acid-reacting, soluble cleavage products.

It would be difficult to understand why the total acids originally present in oats should not dissolve within 1 hour and hence should not be included in the 60-minute acidity value, in view of the fact that, as Thatcher and Koch (*16*) have shown, all of the diastase of cereal products can be brought into solution by a 1-hour extraction with ice water. Diastase, being a colloid, is considered soluble with some difficulty, while acid-reacting substances which occur in nature are known to be readily soluble in water.

From the corn meal acidities recorded in Table IV it would appear that for corn meals the extraction period in the ice-water method should be longer than for oats. For the experiment I used a commercial white corn meal which was sufficiently fine to be passed through a 20-mesh sieve. Notwithstanding this fineness slightly more than 1 hour's time appeared to be required to dissolve with ice water all of the acid contained in this meal. It follows that the extraction period should be extended to 1½ hours for corn possessing this degree of fineness.

On the whole it may be said that the ice-water extraction method affords a very rapid and inexpensive means for determining the original quantity of free acids contained in cereal products, and the method is no doubt of wide applicability. The results obtained by its use are sufficiently accurate for practical purposes. At the same time it permits the experimenter to devise such modifications as may suggest themselves in special cases. For practical routine work the use of a cold-storage room can, of course, be easily dispensed with by immersing the tightly stoppered extraction bottles in a vessel with ice water. In the work of Mr. Mastin, who, as stated, is responsible for the acidity figures recorded in Table II, this mode of operation was employed. It was also found preferable to use a *N*/20 solution of sodium hydroxid for the titrations. In general, the following suggestions for the successful operation of the method may be given:

APPARATUS AND REAGENTS REQUIRED

1. A flat-bottomed pan, with walls at least 12.5 cm. high, which is kept filled with ice water.
2. Extraction bottles of ordinary glass, provided with well-fitting rubber stoppers.
3. Graduated cylinders, capacity 100 cc.
4. Glass funnels.
5. Wide-mouthed beaker flasks, capacity 100 cc.
6. Folded filter paper.

7. A volumetric pipette, capacity 25 cc.
8. A volumetric pipette, capacity 150 cc.
9. An alcoholic solution of phenolphthalein (0.15 to 1 per cent strength).
10. A *N/20* solution of sodium hydroxid.
11. A supply of distilled water, previously freed from carbon dioxid by boiling, kept on hand in the refrigerator or immersed in ice water.

PREPARATION OF SAMPLE

All materials to be tested for acidity should be in a finely divided state in order to make possible a complete and rapid extraction of the acids. It is also to be borne in mind that cereal products usually undergo rapid changes in acidity after they have once been ground.

METHOD OF MAKING THE TEST

Weighed quantities of the finely ground materials, for example 15 gm. of each sample, are placed in the dry extraction bottle, and a tenfold quantity of the precooled distilled water is pipetted into each of the bottles. The latter are now stoppered tightly, well shaken, and immersed in the flat-bottomed pan holding the ice water. At 15-minute intervals they are momentarily removed from the bath to be shaken vigorously. Unless this is done at the prescribed intervals, or oftener, accurate results will not be obtained. While the extractions are in progress preparations may be made for the filtrations. At the end of the extraction period (1 hour for oats, 1½ hours for corn) the mixtures are immediately thrown upon dry folded filters, the filtrates being collected in the graduated cylinders. The first few cubic centimeters of each filtrate may be discarded. If filtration is slow, or during hot weather, it is best to keep the filtrates cool by placing the graduated cylinder in the pan with ice water during filtration. A measured aliquot, for example 25 cc. of each filtrate, is finally pipetted into a beaker flask, and titrated with the alkali, after adding a few drops of phenolphthalein. It has been customary in this country to express the acidity in terms of cubic centimeters of normal alkali required to neutralize the acid extracted from 1,000 gm. of the material. For very accurate work allowance would have to be made in the calculations for the volume occupied by the undissolved portion of the material, but for practical purposes this correction may be omitted.

The titration in the presence of phenolphthalein offers no difficulties, except in cases where the grain product is strongly damaged and discolored. In such cases the filtered extracts may show a deep yellow color, and the endpoint of titration becomes difficult to recognize. This difficulty, if met with, can be overcome by the use of colorimetric devices, such as the apparatus employed by Lüers and Adler (*4,10*).

ACIDITY OF SOUND OATS OF PURE VARIETIES DETERMINED BY THE ICE-WATER EXTRACTION METHOD

It seemed of interest to examine by means of the ice-water method a number of sound oats of known type and origin, and to compare their acidities with those found for the more or less damaged oats listed in Table I. Samples of pure strains of oats were secured from various experiment farms of the Department of Agriculture through the Office of Cereal Investigations, Bureau of Plant Industry. Two samples

received from the individual growers in the central coast region of California and known to be of high quality were included in this experiment. The titration results, calculated to the basis of dry weight, are given in the following table.

TABLE V.—*Acidity values of different varieties of oats shown by the ice-water method*[1]

Sample No.	Variety.	Locality.	Crop year.	Acidity value.
62	Green Russian	Iowa	1914	16.30
63	Silvermine	do	1914	23.00
64	Canadian	Idaho	1915	22.00
65	Silvermine	do	1915	24.00
66	Swedish Select	North Dakota	1915	13.50
67	do	South Dakota	1915	15.75
68	Winter Turf	Virginia	1915	17.50
69	Red Rustproof	do	1915	20.00
70	Sixty Day	South Dakota	1915	13.60
71	Victory	North Dakota	1915	14.00
72	Abundance	do	1915	15.50
73	Siberian	do	1915	19.90
74	Red Oats	California	1914	8.75
75	Black Oats	do	1914	10.50

[1] Expressed in cubic centimeters of normal alkali required to neutralize the extract from 1,000 gm. of material.

If we compare the above acidity values with each other and with those of the unsulphured but damaged oats in Table I, we find a remarkable degree of uniformity, considering that the samples represent oats of two seasons and grown in different localities. We observe, above all, no essential difference in the values for the damaged oats (in Table I) from those of the sound oats (in Table V). This clearly shows that the amount of free acid present in oats does not change materially in the unground kernel during the early stages of spoilage. The lowest acidities found in sound oats were those of the two samples from the Pacific coast, and in no case was the value as high as 25. Incidentally, there are indications that certain constant acidity values are characteristic of the different varieties. Thus the values for samples 66 and 67 in Table V, which are both of the Swedish Select variety and had been grown in different States, are not far apart. Similarly, No. 63 and 65 of the Silvermine variety show nearly the same acidity, although grown in different States and seasons.

SUMMARY

(1) Various deficiencies of the Schindler method for measuring the acidity of cereal products are pointed out. These deficiencies are attributable to the presence of alcohol during the extraction and subsequent titration.

(2) A new and simple method is recommended for determining the amount of free acid originally present in cereal products. The im-

portant feature of the new method is the use of ice water for the extraction of the material.

(3) By means of the ice-water method it is shown that the amount of acid present in oat kernels does not change markedly during the early stages of spoilage. If oats are sulphured their acidity is increased.

(4) Oats which previously had been sulphur bleached showed no increase in acidity upon prolonged standing in the ground state when tested by means of the ice-water method, the acid-forming ferments of the grain having been destroyed by the sulphur fumes. With Schindler's method pronounced increases in the titration values were still observed in these cases, owing doubtless to the fact that certain protein cleavage products continue to be formed, which in aqueous solution are amphoteric, but which possess an acid reaction in the presence of alcohol.

(5) Ice-water extracts of oats or corn, if filtered and kept at the temperature of 1° to 2° C. for 24 hours without being neutralized, undergo no change in acidity. If neutralized, a new formation of acid takes place, notwithstanding the low temperature.

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